

PATENT COOPERATION TREATY

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

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| To: | MONEY | £ |
| DENHOLM, Ann | ORDER | |
| D Young & Co | DIARY | 08 FEB 2005 |
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| GRANDE BRETAGNE | | |
| (LONDON) | | |
| | - 8 NOV 2004 | |
| | ANSO | |
| | ENTRY | |
| | FOR | DS8 LCH |

SOUTHAMPTON

• 8 NOV 2009

PCT

WRITTEN OPINION
(PCT Rule 66)

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| ANS | | Date of mailing (day/month/year) | 08.11.2004 |
| ENTRY | DS LCH | | |
| FOR | | | |
| Applicant's or agent's file reference P013723WO AMD | | REPLY DUE | within 3 month(s) from the above date of mailing |
| International application No. PCT/GB 03/03436 | | International filing date (day/month/year) 06.08.2003 | Priority date (day/month/year) 06.08.2002 |
| International Patent Classification (IPC) or both national classification and IPC C12P19/04 | | | |
| Applicant DANISCO A/S et al. | | | |

1. This written opinion is the **first** drawn up by this International Preliminary Examining Authority.
2. This opinion contains indications relating to the following items:
 - I Basis of the opinion
 - II Priority
 - III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
 - IV Lack of unity of invention
 - V Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
 - VI Certain documents cited
 - VII Certain defects in the international application
 - VIII Certain observations on the international application
3. The applicant is hereby invited to reply to this opinion.

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| When? | See the time limit indicated above. The applicant may, before the expiration of that time limit, request this Authority to grant an extension, see Rule 66.2(d). |
| How? | By submitting a written reply, accompanied, where appropriate, by amendments, according to Rule 66.3. For the form and the language of the amendments, see Rules 66.8 and 66.9. |
| Also: | For an additional opportunity to submit amendments, see Rule 66.4. For the examiner's obligation to consider amendments and/or arguments, see Rule 66.4 bis. For an informal communication with the examiner, see Rule 66.6. |
4. If no reply is filed, the international preliminary examination report will be established on the basis of this opinion.
4. The final date by which the international preliminary examination report must be established according to Rule 69.2 is: 06.12.2004

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| <p>Name and mailing address of the international preliminary examining authority:</p> <p> European Patent Office - P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk - Pays Bas Tel. +31 70 340 - 2040 Tx: 31 651 epo nl Fax: +31 70 340 - 3016</p> | <p>Authorized Officer Smalt, R Formalities officer (incl. extension of time limits) Humbert, C Telephone No. +31 70 340-4129</p> |
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I. Basis of the opinion

1. With regard to the **elements** of the international application (*Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this opinion as "originally filed"*):

Description, Pages

1-59 as originally filed

Claims, Numbers

1-43 as originally filed

Drawings, Sheets

1/20-20/20 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
- the language of publication of the international application (under Rule 48.3(b)).
- the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- contained in the international application in written form.
- filed together with the international application in computer readable form.
- furnished subsequently to this Authority in written form.
- furnished subsequently to this Authority in computer readable form.
- The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
- The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- the description, pages:
- the claims, Nos.:
- the drawings, sheets:

5. This opinion has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

6. Additional observations, if necessary:

V. Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N) Claims 1-6,9-43

Inventive step (IS) Claims 1-43

Industrial applicability (IA) Claims

2. Citations and explanations

see separate sheet

1. The following documents (D) are referred to in this communication; the numbering will be adhered to in the rest of the procedure:

- ✓ D1: WO 94/12656 A (QUEST INT ;DEN BERG DIRK JOHANNES CORNELI (NL); LEDEBOER ADRIANUS) 9 June 1994 (1994-06-09)
- ✓ D2: WO 01/57234 A (AHLGREN JEFFREY A ;SANDINE WILLIAM E (US); KNOSHAUG ERIC P (US); O) 9 August 2001 (2001-08-09)
- ✓ D3: VAN GEEL-SCHUTTEN, G H ET AL.: "Biochemical and structural characterization of the glucan and fructan exopolysaccharides synthesized by Lactobacillus reuteri wild-type and mutant strains" APPLIED AND ENVIRONMENTAL MICROBIOLOGY, WASHINGTON,DC, US, vol. 65, 1999, pages 53-72, XP002191551 ISSN: 0099-2240
- ✓ D4: VAN HIJUM, S.A.F.T. ET AL.: "Molecular characterization of a novel fructosyltransferase from Lactobacillus reuteri synthesizing a high molecular weight fructan with beta-(2->1) linked fructosyl units in Escherichia coli" APPLIED AND ENVIRONMENTAL MICROBIOLOGY, WASHINGTON,DC, US, vol. 65, 1999, pages 73-95, XP002264246 ISSN: 0099-2240
- ✓ D5: US-A-4 444 793 (BODIE ELIZABETH A ET AL) 24 April 1984 (1984-04-24)
- ✓ D6: EP-A-0 881 283 (NESTLE SA) 2 December 1998 (1998-12-02)
- ✓ D7: WO 00/47727 A (MAX PLANCK GESELLSCHAFT ; KOSSMANN JENS (DE); KNUTH KAROLA (DE); QUANZ) 17 August 2000 (2000-08-17)
- ✓ D8: WO 03/008618 A (TNO ;VAN GEEL-SCHUTTEN GERRITDINA H (NL)) 30 January 2003 (2003-01-30)
- ✓ D9: ZAHNLEY J C ET AL: "Glycosyltransferase profiles of representative strains of Leuconostoc mesenteroides" FASEB JOURNAL, vol. 11, no. 9, 1997, page A1420, XP001160862 17th International Congress of Biochemistry and Molecular Biology in conjunction with the Annual Meeting; San Francisco, California, USA; August 24-29, 1997 ISSN: 0892-6638

Re: V

2. Novelty

2.1 In D1 a *Lactobacillus sake* strain is described, which produces an EPS *in situ* in various food products, including milk. The enzymes involved in the formation of this EPS

are not characterized, but the EPS is shown to comprise glucose and rhamnose units. D1 does not mention pharmaceutical applications. This document therefore takes away the novelty in the sense of Art.33(2) PCT of claims 1-6,9,15-21,23-29,35,37-43.

2.2 D2 describes a *Lactobacillus lactis* strain "Ropy 352", which encompasses a plasmid encoding 13 enzymes involved in the biosynthesis of an EPS consisting mainly of glucose and galactose. The ORF's of the enzyme-encoding plasmid is given for the 13 enzymes, but they are not further characterized. The strain is applied in milk products for EPS production *in situ*, and pharmaceutical applications are contemplated. D2 is hence considered prejudicial to the novelty of claims 1-4,9,15-21,23-34,37-43 in accordance with Art.33(2) PCT.

2.3 D3 describes in table 1 homo-EPS of 100% glucose and also of 100% fructose, made by a *Lactobacillus reuteri* strain, which expresses glucan sucrase and secretes it into the culture medium. In view thereof, claims 1-4,9,15-21,23-29,33,37-43 are not new over D3 in the sense of Art.33(2) PCT.

2.4 In D5 the use of *Leuconostoc mesenteroides* in a fermented whey product is described. The organism produced a dextran *in situ* having thickening, stabilizing, and emulsifying properties, through a dextran sucrase activity (a glycosyl transferase; see also D9). Spray-drying of the broth is contemplated/ suggested; see last sentence of example 2. D5 is therefore prejudicial to the novelty of claims 1-3,10-17 and 19-43.

2.5 D6 discloses the use of *Leuconostoc mesenteroides* for producing an EPS with nutritional and pharmaceutical application, though the use of its dextran sucrase activity. Here it is explicitly used in milk fermentation. Present claims 1-3 and 10-43 are therefore not new over D6 in the sense of Art.33(2) PCT.

2.7 In D7 the cloning of alternansucrase as well as dextran sucrase from *Leuconostoc mesenteroides* is described (see examples 3 and 4), as well as the use in food and cosmetics of the EPS produced by these enzymes (homoglucans). D7 therefore takes away the novelty in accordance with Art.33(2) PCT of present claims 1-3,10-17,21,35-37 and 39.

2.8 D8 is a PCT application published between the priority and the filing date of the

present application, but with priorities which precede the priority date of the present application. In the international phase under the PCT, such a document is not considered to be comprised in the prior art in accordance with Rule 33.1 PCT, unless the priority is not validly claimed, which in this case seems to be in order. No unified criteria exist for the interpretation of such documents in the various national and regional phases under the PCT. Under the EPC, D8 is considered as part of the state of the art for novelty purposes only, since it has validly entered the regional phase, to the extent that overlapping EPC contracting states have been validly designated. D8 describes *Lactobacillus sake* glucansucrase/ glucosyltransferase and *Leuconostoc* glucansucrase, glucans produced thereby, and their use in food and pharmaceutical products, including *in situ* synthesis in yoghurt. D8 is therefore anticipated to be relevant for present claims 1-10,12-21,23-32,35-39 and 41-43.

3. Inventive step

3.1 The additional technical features of claims 22 and 36 are natural extrapolations of the teaching of D1, and cannot be considered to contribute an inventive step in accordance with Art.33 (3) PCT.

3.2 It is common general knowledge in the technical field of this application that many lactic acid bacteria produce various useful EPS's, and that different combinations of a limited number of the same or similar enzymes are involved in the synthesis thereof. The difference between the subject-matter of claims 5-8, 35 and 36 and e.g. D1 is that the same or similar products have been found to be synthesized by the same group of enzymes in different isolates, species or subspecies of *Lactobacillus*. The problem solved by the application is therefore the provision of alternative isolates or species of *Lactobacillus* to produce the claimed EPS. The skilled person is continuously searching for such alternative isolates, as is obvious from the body of prior art cited in the search report; that in itself is obvious and thus not inventive in the sense of Art.33(3) EPC. The manner in which the EPS's themselves are described in both the application and the prior art does not allow to distinguish between them. However, should they be different in any aspect, they must be considered as obvious alternatives, unless an unexpected useful property can be demonstrated. In the absence thereof, the species or isolate of the lactic acid bacterium, the EPS, nor the method of use and/or production can be considered to involve an inventive step in the sense of Art.33(3) PCT. This objection affects all claims presently

on file, with the exception of claims 10-12, relating to *Leuconostoc*.

3.3 In D4, the cloning of a fructosyl transferase from *Lactobacillus reuteri* is described. The enzyme is capable of forming a homo-EPS comprising exclusively fructose residues. The organism is food-grade (GRAS), and use of the organism and/or the enzyme and/or the fructan in food (e.g. probiotic, emulsifier, gelling agent) and non-food applications (e.g. anti-cancer, anti-ulcer) is clearly suggested; see the first two paragraphs of the introduction. The core of the present invention, albeit with a different species of *Lactobacillus*, is therefore explicitly suggested by D4. Claims 1-4, 9, 13-34 and 37-43 are hence not considered as inventive in the sense of Art.33(3) PCT.

3.4 The above paragraphs of D4 also mention that many lactic acid bacteria are known to produce an abundant variety of useful homo- and heteropolysaccharides. In analogy with § 3.2 above, such a statement is considered a clear incentive to investigate or test other lactic acid bacteria for their usefulness in this context, in particular in respect of their fructosyl transferase activity. Present claims 5-8, 35 and 36, which relate to specific isolates or (sub)species of *Lactobacillus*, EPS's produced thereby, and their use, can therefore not be regarded as inventive in the sense of Art.33(3) PCT, unless some unexpected useful property can be attributed to them. Such property appears not to be provided by the present application.

3.5 Claim 18 relates to fermenting milk, whereas D5 relates to fermentation of whey. The technical effect of this fermentation, the introduction of EPS, is the same, and in the context of this technical field, milk would be a natural extrapolation from whey (see also e.g. D6), for which no inventive step in accordance with Art.33(3) PCT can be recognized.

4. Conclusion

All claims presently on file have either novelty or inventive step problems. It would appear that the EPS's of the application cannot be characterized in a manner which would make them new *per se*. The specific EPS-producing strains as deposited are new, as is their use and compositions comprising them. Claims restricted to such subject-matter can however only be considered to involve an inventive step if the organism, the process, the use, or the composition comprising the organism has an unexpected technical feature. Such features are not at present apparent from the application. Should the applicant consider that there

is patentable subject-matter present in the application, he or she is invited to file new claims, taking into account the above objections. The applicant is in that case invited to indicate where basis for such claims can be found, in what manner they differ from the previous claims, and how that is considered to affect patentability.